

THE CHEMICAL INVESTIGATION OF SOME POISON-
OUS PLANTS IN THE N.O. SOLANACEÆ.

PART II. *NICOTIANA SUAVEOLENS*, AND THE IDENTIFICATION
OF ITS ALKALOID.

BY JAMES M. PETRIE, D.Sc., F.I.C., LINNEAN MACLEAY FELLOW
OF THE SOCIETY IN BIOCHEMISTRY.

(From the Physiological Laboratory of the University of Sydney.)

Nicotiana suaveolens Lehm., the "native tobacco" of Australia, and the only endemic species, is plentiful in the interior of this State. It grows about three feet high, and is often a troublesome weed in the stock country. It is a drought-resistant plant, and spreads over large tracts of land in the dry seasons. Hence it is that, when grass and other fodder plants are withered or overrun by this weed, it is often the only green plant left available to starving animals. It is then readily eaten by stock, and, according to the reports of the owners and Inspectors, the results are variable. Though in many cases no apparent harm has followed, there is still a consensus of opinion among stockmen, that many of their losses must be attributed to this plant.

The only record of tests having been made on this species, is a paper by Dr. Bancroft (Proc. Roy. Soc. Queens., iv., 1887, p.9), in which he states that the physiological effect of the extracts on animals resembled that of extracts of true tobacco and of pituri.

The following is the account of a chemical investigation of this plant, which was undertaken to decide definitely the nature of its active principle, and also to determine whether this constituent is present in quantity sufficient to cause death.

Extraction of active principle:—For this purpose, plants were collected in the midsummers of 1911, 1912, and 1913, chiefly from the dry North-West. Through Chief Inspector Symons, of the Stock Department, a sample was received from Narrabri. This, on its arrival, contained 37 per cent. of moisture, and consisted of leaves, stalks, and roots. The whole sample was extracted with alcohol, and the solvent afterwards removed by

distillation under diminished pressure. The extract gave all the general alkaloid reactions, and smelt strongly of tobacco. The alkaloid was completely removed from this extract by petroleum spirit (b.p. under 45°C) after making alkaline with sodium hydroxide. From this coloured solution, the alkaloid was carefully purified without loss, by shaking it into water and petroleum spirit successively, many times, and finally obtained as a colourless, aqueous solution. This solution was slightly alkaline, and possessed the odour of nicotine. It was then titrated with tenth-normal acid and alkali, and gave an equivalent of 2 c.c. of acid neutralised by the alkaloid. If this quantity be calculated as nicotine, it represents 0.0324 gm., and is 0.124 per cent. of the plant (dried at 100°).

A second sample, from the Castlereagh River, in the Coonamble district, was obtained from Mr. Breakwell, of the Department of Agriculture. This sample had been spread out to dry in the air to avoid mould in transit, and when received it contained only 9 per cent. of moisture. The whole of the material, consisting of leaves and stalks, in this case was subjected to distillation in a current of steam, the powdered plant being first mixed with 0.5 per cent. sodium hydroxide in solution, and a large excess of milk of lime. The whole of the alkaloid passed into the distillate; and the residue in the still being free from alkaloid, showed that no non-volatile alkaloid existed in the plant. The voluminous distillate contained much ammonia, which is derived from the cleavage of amido compounds, and this free ammonia was eliminated by passing a current of air through the solution for many hours. The alkaloid was next converted into oxalate, and the fluid concentrated at a low temperature to about 300 c.c. From this solution ether removed the alkaloid, and the ether extract was carefully purified and dried. The ether was then slowly removed, and the residue dried to constant weight; 0.07 gm. was obtained, which represented 0.011 per cent. of the plant-material dried at 100°C .

Another quantity was collected for me, near Picton, about 50 miles from Sydney, by Mr. E. Cheel, of the National Herbarium. This consisted of fresh, green leaves and stalks, with 72 per cent.

of moisture. The whole was distilled as before, in a current of steam, until the alkaloid was completely volatilised. The alkaloid was isolated and purified as in the last case, neutralised with excess of tenth-normal oxalic acid, and the excess determined by titration, using cochineal indicator. The result in this case gave 0.0178 gm. of alkaloid, or 0.015 per cent. of the plant (dried at 100°).

If we regard the above sample containing 72 per cent. of water as a fair average specimen of fresh material, we can express these results also in terms of the green plant, and thereby form a better idea of the amount of alkaloid in the original plant as eaten by stock. The results may then be stated:—

Amount of Alkaloid expressed as Nicotine.

(1) 0.035% of fresh plant.	0.124% of plant dried at 100°C.
(2) 0.003% of fresh plant.	0.011% of plant dried at 100°C.
(3) 0.004% of fresh plant.	0.015% of plant dried at 100°C.

Examination of the Alkaloid,—The aqueous solution is alkaline to litmus, and possesses a burning taste, and the characteristic tobacco odour. The pale yellow substance, when exposed to the air, oxidises, and turns dark brown; it then possesses the nauseating odour of nicotine.

Of the salts of nicotine, the most characteristic, and the one best adapted for the identification of the alkaloid, is the picrate. Accordingly, the picrate was prepared from the aqueous solution by the addition of excess of picric acid. The dense yellow precipitate, amorphous at first, gradually assumed, on standing, the characteristic, thin, yellow, needle-shaped crystals. At the same time, pure nicotine picrate was prepared under similar conditions, and the crystals compared. Under the microscope, they were precisely alike. The crystals were washed completely with distilled water, and recrystallised three times from water, then finally dried at 100°C. The melting-points were then determined together:—

Picrate of <i>N. suaveolens</i> alkaloid	—m.p. 218°C.(corrected).
Picrate of pure nicotine.....	218
The two mixed together.....	218

The melting-point of nicotine picrate has been determined by Pinner and Wolffenstein as 218°C.(Ber.24, 1891, 66).

The alkaloid of *Nicotiana suaveolens* is therefore nicotine.

For the purpose of comparison, pure nicotine tartrate (B.W.) was decomposed, and the nicotine distilled from it in a current of purified hydrogen gas. It was collected and at once sealed up in the receivers. From this colourless liquid, the nicotine picrate was prepared.

Toxicity of the plant:—Nicotine is probably the most violent poison known. Wynter Blyth gives the lethal dose for a human adult as about 6 mgs. In Abderhalden's "Biochemisches Handlexikon," it is stated that 5 mgs. suffice to kill a medium-sized dog in three minutes. It is evident from these data, taking even the lowest value of nicotine in the above results, that there is enough contained in one half pound of the green plant, to poison an ordinary sized sheep.

References to the plant as a stock poison:—Of the 80 or more species of *Nicotiana*, only a few are known to contain nicotine. *Nicotiana suaveolens* being limited to the Australian continent, the records of fatalities are all local. But it is referred to, also, by European authorities, such as Dragendorff in "Die Heilpflanzen," (1898) as a poisonous plant; by Greshoff in his "Monographia de plantis venenatis" as poisonous for cattle; and by Pammel ("Poisonous Plants," 1911) as poisonous to stock. It is described by F. M. Bailey, as a stock poison in Queensland, and by Professor Ewart as a feebly poisonous plant in Victoria. Mr. J. H. Maiden states that it is very deadly to all stock, and refers to many instances of poisoning of cattle, sheep, pigs, and rabbits. In his "Plants reputed poisonous to Stock," Mr. Maiden describes a sudden fatality, in 1891, of 300 healthy cattle, travelling on the great stock route through Milparinka.

Summary.—The results of this paper prove that *Nicotiana suaveolens* contains the extremely poisonous alkaloid nicotine, and that the nicotine is present in sufficient quantity to poison stock.

I express my thanks to Professor Sir Thomas Anderson Stuart, in whose laboratory the work was done.